Soil and Plant Factors Influencing the Accumulation of Heavy Metals by Plants

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The use of plants to monitor heavy metal pollution in the terrestrial environment must be based on a cognizance of the complicated, integrated effects of pollutant source and soil-plant variables. To be detectable in plants, pollutant sources must significantly increase the plant available metal concentration in soil. The major factor governing metal availability to plants in soils is the solubility of the metal associated with the solid phase, since in order for root uptake to occur, a soluble species must exist adjacent to the root membrane for some finite period. The rate of release and form of this soluble species will have a strong influence on the rate and extent of uptake and, perhaps, mobility and toxicity in the plant and consuming animals. The factors influencing solubility and form of available metal species in soil vary widely geographically and include the concentration and chemical form of the element entering soil, soil properties (endogenous metal concentration, mineralogy, particle size distribution), and soil processes (e.g., mineral weathering, microbial activity), as these influence the kinetics of sorption reactions, metal concentration in solution and the form of soluble and insoluble chemical species.

The plant root represents the first barrier to the selective accumulation of ions present in soil solution. Uptake and kinetic data for nutrient ions and chemically related nonnutrient analogs suggest that metabolic processes associated with root absorption of nutrients regulate both the affinity and rate of absorption of specific nonnutrient ions. Detailed kinetic studies of Ni, Cd, and Tl uptake by intact plants demonstrate multiphasic root absorption processes over a broad concentration range, and the use of transport mechanisms in place for the nutrient ions Cu, Zn, and K. Advantages and limitations of higher plants as indicators of increased levels of metal pollution are discussed in terms of these soil and plant phenomena.

The principal objectives of this review are to briefly describe the soil and plant factors influencing trace metal uptake by plants, and, with this information as a basis, illustrate some of the parameters which must be considered in using higher plants as indicators of increased levels of metals in the terrestrial environment.

In order to utilize plants as monitors of metal pollution in the field, it is necessary to distinguish between uptake arising from natural metal sources and from pollutant sources. Metals from both natural and pollutant sources have the potential for being assimilated by the plant through foliar or root absorption processes. The importance of foliar absorption processes for several heavy elements has been discussed elsewhere (*1, 2*). Separation of the sources of metals taken up by roots is complicated by the mediating effect of soil properties and soil and plant processes. These effects may be illus-

trated by examination of the concentration of metals in soil relative to possible increases due to pollutant sources and the soil and plant factors influencing plant accumulation of metals.

Soil Factors Influencing Plant Uptake of Metals

Soil Metal Concentration

Soils represent the major repository of trace elements over geologic time. On a worldwide basis, soils exhibit an average composition close to the earth crust (Table 1), but the near-surface parent material from which soils are derived is not uniform and soil-forming processes differ markedly from one climatic region to another, accounting for considerable overall variability in trace metal concentrations (*5*). Increases in average trace element levels estimated to arise from the deposition of fly ash from a 1400 MWe coal combustion plant

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Table 1. Concentration of selected elements in soils on a worldwide basis and projected increases due to coal combustion.

Element[a]	Concentration in soil[a]		Estimated increase in soil concentration due to coal combustion[b]	
	Concentration range, μg/g	Average concentration, μg/g	μg/g	%
Ag	0.01-5	0.1	3×10^{-5}	3×10^{-2}
As	0.1-40	6.0	1×10^{-3}	2×10^{-2}
Be	0.1-40	6.0	3×10^{-4}	5×10^{-3}
Cd	0.01-0.7	0.06	1×10^{-4}	2×10^{-1}
Co	1-40	8	1×10^{-4}	2×10^{-3}
Cr	5-3000	100	8×10^{-3}	8×10^{-5}
Cu	2-100	20	1×10^{-2}	6×10^{-2}
Ga	0.4-300	30	3×10^{-3}	1×10^{-2}
Ge	1-5		8×10^{-3}	8×10^{-1}
Hg	0.01-0.8	0.03	2×10^{-4}	7×10^{-1}
Mn	100-4000	850	1×10^{-2}	1×10^{-3}
Mo	0.2-5	2	2×10^{-3}	1×10^{-1}
Ni	10-1000	40	5×10^{-3}	1×10^{-2}
Pb	2-200	10	2×10^{-3}	2×10^{-2}
Sb	NA	1 (lithosphere)	2×10^{-3}	2×10^{-2}
Se	0.01-2	0.2	2×10^{-3}	1×10^{-2}
Sn	2-200	10	4×10^{-3}	4×10^{-2}
Tl	NA	0.3 (lithosphere)	3×10^{-5}	1×10^{-2}
Th	0.1-12	5	4×10^{-4}	8×10^{-3}
V	20-500	100	2×10^{-3}	2×10^{-3}
W	NA	1 (lithosphere)	3×10^{-3}	3×10^{-1}
Zn	10-300	50	1×10^{-2}	3×10^{-2}

[a] Data of Bowen (3) and Swaine (4).

[b] Increased concentration assumes soil depth of 20 cm and soil density of 1.6 g soil/cm³ and is based on estimated (1) 40 year accumulation of fly ash from a 1400 MWe power plant in the area of maximum deposition.

[c] NA = not available.

operating with electrostatic precipitators over a 40 year period (1) are relatively low, amounting to less than 1% (Table 1). Increases in most of the elements amount to less than 0.1% of the total with only Cd, Ge, Hg, Mo, Se, and W meeting or exceeding this value. Estimated trace element deposition from a coal combustion plant without electrostatic precipitators (6) was several orders of magnitude greater than reported in the above study. However, the increases in soil concentration estimated by either study were generally well within the range of variability between soils on a world-wide basis and often on a regional basis. Thus, measurement of changes in the total concentration of these elements in soil, would be an uncertain indicator of increases in environmental levels due to coal combustion. There is, therefore, a need to develop other chemical or biological measures of increased levels and plant availability of trace metals in soil from this source.
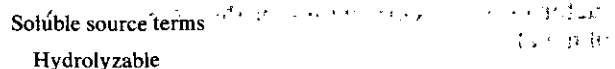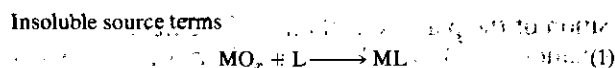
In addition to fossil fuel combustion, trace elements may enter soil indirectly as a result of industrial activity and, directly, in municipal wastes (7, 8) fertilizers, or other soil additives (9). Several of these sources, or disposal of stack scrubber wastes, may result in locally higher concentrations of trace

elements in soils than estimated to arise from releases to the atmosphere and increases may be detectable by chemical analysis. However, a key consideration in use of plants as monitors of these increases will be the availability of the elements for plant uptake.
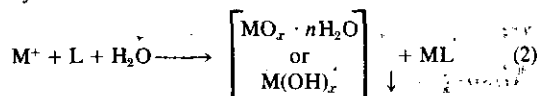
Soil Processes and Properties

The major factor governing availability to plants in soils likely will be the solubility and the thermodynamic activity of the uncomplexed ion (10) since in order for root uptake to occur, a soluble species must exist adjacent to the root membrane for some finite period. The form of this soluble species will have a strong influence on its longevity in soil solution, mobility in soils, and on the rate and extent of uptake, and perhaps, mobility and toxicity in the plant (11).

Once deposited, metal-containing materials are subject to chemical and microbial modification with metal solubility ultimately approaching thermodynamic equilibrium with native soil minerals and organic matter. The rate and extent of solubilization are governed by the physicochemical properties of the deposited material, soil processes, and soil properties.
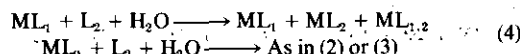
Insoluble source terms

$$MO_x + L \longrightarrow ML \quad \text{(1)}$$

Soluble source terms

Hydrolyzable

$$M^+ + L + H_2O \longrightarrow \begin{bmatrix} MO_x \cdot nH_2O \\ \text{or} \\ M(OH)_x \end{bmatrix} + ML \quad \text{(2)}$$

Nonhydrolyzable

$$M^+ + L + H_2O \longrightarrow MO_x^+ + ML \quad \text{(3)}$$

Organic complexes

$$ML_1 + L_2 + H_2O \longrightarrow ML_1 + ML_2 + ML_{1,2}$$
$$ML_1 + L_2 + H_2O \longrightarrow \text{As in (2) or (3)} \quad \text{(4)}$$

The major sources of metals to the soil may be classified according to expected initial solubility in soil [Eqs. (1)-(4)], where metals are represented by M and L represents organic and inorganic ligands capable of reacting with metals and forming soluble or insoluble products (12). Particulate oxides such as those arising from fossil fuel combustion or nuclear fuel reprocessing initially may be expected to be largely insoluble in the soil solution. Ultimately, solubility should be a function of the composition, configuration, and equivalent diameter of the particle as well as soil properties and processes. Oxide particles containing the highest concentrations of impurities in the crystal lattice may exhibit greatest solubility. The combination of configuration and equivalent diameter as reflected in surface area exposed to solution will be the other main factor governing oxide solubility. Once solubilized, the metals will be subject to the chemical reactions governing soluble salts. Hydrolyzable metals (e.g., Ni, Cd) or metals forming insoluble precipitates with S or P on entering the soil in soluble forms may be expected to be rapidly insolubilized at the near neutral pH of most soils due to hydrolysis, on dilution and subsequent precipitation on, or reaction with, particle surfaces (13). Certain elements (e.g., Fe) may also form precipitates with S or P (14, 15). Conversely, metals not subject to marked hydrolysis (e.g., Tl) may be initially more soluble. Metals with low ionic potentials tend to form primarily simple soluble ions while metals with intermediate and high ionic potentials tend to form soluble complexes. Common inorganic complex-forming ions in the soil solution include CO_3^{-2}, HCO_3^-, SO_4^{-2}, S^{-2}, HS^-, OH^-, and Cl^-. Recent evidence (16, 17) indicates that soil microorganisms may play an important role in this process through the production of soluble ligands with high affinity for metals. Information on organic ligands in solution is limited but both inorganic and organic complexation may result in orders-of-

magnitude solubility increases, above those concentrations predicted by solubility product calculations, for the most stable solid phase. Immobilization of those elements not subject to hydrolysis and precipitation may occur through ion exchange reactions with particulate surfaces. Solubility in soil may be particularly pronounced if the species stable in aqueous solution are anions (e.g., Se, W) that are not tightly bound by the predominantly negatively charged soil. Some ion selectivity occurs through specific chelation with organic matter or interaction with inorganic surfaces and is generally a function of ion valence and hydrated radius. Selectivity usually decreases with increasing valence and increases with decreasing hydrated ion size. Complicating this situation, disproportionation and complexation reactions may occur concurrently, depending upon the element.

Metals entering the soil as stable organo-complexes, such as those used in fertilization to correct micronutrient deficiencies or those possibly present in discharge from a nuclear fuel separation facility, may be initially highly soluble (18, 19). The duration of solubility and mobility in the soil will be a function of the stability of the complex to substitution by major competing ions, such as Ca and H (20-22) and the stability of the organic ligand to microbial decomposition (19). The disruption of the complex may lead to marked reduction in metal solubility through hydrolysis, precipitation or exchange reactions as described above. A portion of the ion released may react with other, perhaps more stable, ligands in soil. The mobility of the stable intact complexes, in turn, will be principally a function of the charge on the complex which will govern the degree of sorption on soil particulates.

Further generalizations of metal behavior on the basis of source term are complicated by the overwhelming importance of soil properties and processes in influencing metal behavior on a regional and local basis. Soil physiocochemical properties may be expected to have complex, interdependent effects on metal solubility. On the basis of research with trace metals exhibiting a range in chemistries, it may be concluded that the soil physicochemical parameters most important in influencing the solubility of metals include: solution composition (inorganic and organic solubles), Eh, and pH; type and density of charge on soil colloids; and reactive surface area (15). These phenomena will be dependent upon soil properties including metal concentration and form, particle size distribution, quantity and reactivity of hydrous oxides, mineralogy, degree of aeration and microbial activity (10, 15, 17). These soil properties are highly variable geographically and will be a function of the combined effects of

parent material, topography, climate, biological processes, time and man's activities (23, 24). It is clear that the soil factors influencing the concentration, form, and plant availability of metals are highly complex, and use of plants as monitors of increased metal levels arising from pollutant sources is dependent upon a detailed understanding of influential phenomena for specific pollutants, soils, and geographical locations.

Plant Factors Influencing Metal Uptake

The myriad of parameters regulating the chemical fate of specific elements in soils determine their solubility and availability for plant uptake. The plant uptake of chemical species in soil solution is also dependent on a number of plant factors. These include: physical processes such as root intrusion, water, and ion fluxes and their relationship to the kinetics of metal solubilization in soils; biological parameters, including kinetics of membrane transport, ion interactions, and metabolic fate of absorbed ions; and the ability of plants to adapt metabolically to changing metal stresses in the environment.

Physical Aspects of Ion Replenishment in the Rhizosphere

The relative efficiency with which plants harvest both essential nutrients and nonnutrients from soil is dependent in part on the interrelationships between plant and soil physical factors. The process of plant root intrusion within the soil profile provides an extensive rhizosphere for ion absorption. Dittmer (25) has shown that after 4 months of growth in a 0.052 m³ container of soil, the roots of winter rye had a surface area of 639 m² and a combined length of 623 km. Although this provides an effective absorptive surface in contact with soil particles and associated soil solution, the concentration of individual ions in solution can be rapidly depleted by plant uptake. Depletion of ions in the rhizosphere is alleviated to some extent by diffusion of ions, and by mass flow of both water and ions from surrounding soil induced by transpirational demand of the plant (26). Ultimately, the supply of ions within the rhizosphere is controlled by the kinetics of solubilization of ions sorbed to the solid phase of soil, as discussed above, and the kinetics of removal by the plant root. Barber and Claassen (27, 28) have developed mathematical models to describe metal uptake by plants based on the above kinetic parameters. In effect, the elemental compo-

sition of the plant reflects to some extent, the composition of the soil solution; this represents a key factor in our understanding of the uptake behavior of metals.

Kinetic Parameters Regulating Plant Absorption of Metals

Over the past several years the authors and coinvestigators have studied the behavior of 15 trace metals in plants. These studies have indicated that: abiotic and biotic soil processes controlled the solubility and availability of metals for plant uptake; metals were taken up by plants at differing rates; and metals, once absorbed, varied as to their mobility within the plant, suggesting a second point of metabolic regulation. The complexities involved in attempting to employ plants as indicators of environmental pollution are illustrated by the results of investigations to compare the bioavailability of a number of endogenous soil elements and soluble amended metals (Table 2). It should be noted that the reported concentration ratios (CR values) are based on the total endogenous soil concentration of each element and on total metal amended (2.5 ppm). Only a small fraction of the endogenous metal is soluble and therefore available; similarly, although amended metals were supplied in soluble forms, solubility of nonvolatiles ranged from <1% (Pb) to 40% (Tl) after 13 days incubation in soil. Therefore uptake as indicated by the CR values reported are low for both amended and endogenous elements. Elements comprising both the endogenous and amended metal pools are accumulated by the plants. Calculated CR values, based on these two pools, indicate that the amended metals were more available than their corresponding endogenous counterparts for 10 of the 15 metals studied. This could have been anticipated since they were supplied initially as soluble salts. However, five of the elements amended to soil, namely, Ag, Cd, Hg, Sn, and Tl, exhibit lower CR values than metals arising from the corresponding endogenous pools. This would suggest that these elements persist as stable, soluble species within the soil solution and are more available for plant uptake. This may result from differences in the nature of metal association with the solid phase or in soluble species, which influence the type and kinetics of exchange between endogenous metals and added metals. It is apparent that, as discussed earlier, the source of metals and time play major roles in influencing plant availability in soil. The implication in terms of the use of plants as monitors of metal pollution is clear, i.e., uptake may not properly reflect recent influxes of metals into the environment.

Table 2. Comparison of availability of endogenous and amended soil metals for uptake by soybean (*Glycine max*) plants grown (60 days) in 400 g Ritzville silt-loam with (2.5 μg/g) and without added metals.

Element[a]	Endogenous soil concentration, μg/g	Plant concentration resulting from endogenous metal, μg/g	Plant concentration resulting from amended metal, μg/g	Concentration ratios[b]		Short distribution at maturity[d]
				Endogenous metal	Amended metal[c]	
Ag	1	0.057	0.003	0.057	0.001 (9.4)	S>>L=P
As	4.6	0.217	1.053	0.047	0.421 (8.8)	—
Cd	0.30	0.301	2.01	1.00	0.804 (34)	L>S>P
Co	34	<0.06	0.398	<0.002	0.159 (10)	—
Cr	59	0.095	0.150	0.002	0.060 (9.5)	S>L>P
Cu	31	3.90	1.70	0.126	0.680 (3.3)	—
Hg	0.017	0.20	0.10	11.8	0.04 (0.8)	L>S>P
Mn	720	26.1	26.4	0.036	10.6 (37)	—
Mo	<5	3.7	476.3	>0.74	190 (9.1)	—
Ni	28	0.62	14.3	0.022	5.72 (20)	P>S>L
Pb	5	5.1	24.6	1.02	9.84 (<1)	—
Sb	0.48	0.0045	0.31	0.009	0.124 (8.9)	L>S>P
Sn	<1	1.1	0.96	>1.10	0.384 (<20)	—
Tl	0.33	0.44	1.47	1.33	0.588 (40)	L>S>P
Zn	74	12.1	10.4	0.164	4.16 (8.2)	—

[a] Metal analysis by atomic absorption spectroscopy and radioanalysis.

[b] Concentration ratio = μg of metal/g of oven-dry plant tissue per μg metal/g of oven-dry soil.

[c] Values in parentheses indicate solubility of soil amended metals after 13 day incubation.

[d] Based on tissue concentration; stem = S, leaves = L, seeds and pods = P.

A comparison of CR values for amended metals (Table 2), indicates a broad range in plant availability, especially considering the proportion of amended metal which is soluble. Relatively higher CR values are not only obtained for nutrient species (Mo, Mn, Cu, and Zn) but also for nonnutrient species such as Pb, Ni, Cd, Tl, As, and Sn. This suggests that the uptake of nonnutrient elements may be metabolically facilitated.

Essential nutrients exhibit two types of distribution in shoot tissues: relatively uniform distribution with leaves being the major site of deposition and transport within the plant through passive movement in the xylem and initial uniform shoot distribution, with remobilization of specific elements from leaves through phloem transport during senescence, to either developing leaves and/or seeds. The distribution of Cd, Hg, Sn, and Tl (Table 2) is similar to nutrient species such as Mg, K, Cu, and Mo, reflecting the potential for remobilization from senescing tissues. Nickel is readily remobilized *from senescing tissues and accumulated in seeds*; a tendency which is shared by a number of nutrilites, i.e., Fe, Cu, Mn, and Zn. Although not as obvious as Ni, Cd also exhibits a tendency for accumulation in seeds of soybean. The elements Ag and Cr are not very mobile within the plant and accumulate in lower stems. Since distributions of specific ions may vary, tissue selection becomes important when employing plants as monitor systems.

Complexation of ions may be the physiological mechanism responsible for the mobility of ions in

the plant (*11*). In the case of nutrient species, Ca, Fe, Cu, Mn, and Zn can be shown to exist within the plant as organometallic complexes. Complexation may provide a basis for maintaining the solubility and mobility of chemically reactive species, permit conservation of substrates by allowing for remobilization, and provide a means of compartmentalization. In addition, complexation of nonnutrients may represent a mechanism for detoxification. This aspect will be addressed in conjunction with accumulator or indicator species.

Although it is generally conceded that the uptake of nutrilites by plants is metabolically regulated, there is some question as to the mechanisms controlling the absorption of nonnutrient species. Much of the information required to understand the behavior of metal pollutants in plants can be extrapolated from an extensive data base available for nutrient species. Peculiar to higher plants is a pattern of ion uptake referred to as multiphasic uptake. Basically, as the concentration of an ion surrounding the root is increased, uptake or absorption by the plant exhibits a series of distinct isotherms, each of which has different kinetic characteristics. These kinetic constants, K_m and V_{max}, describe the affinity of the transport mechanism for a given ion and also the rate of uptake at half-saturating ion concentrations.

Table 3 lists kinetic constants for the transport of a number of nutrient species. Based on these data, several points can be made concerning the uptake potential of plants. First, each ion exhibits a number

Table 3. Kinetic constants for absorption of nutrient ions.

Ion	Phase	K_m, μM	V_{max}, μmole/2 dry wt root-hr	Plant tissue	Ref.
K^+	2	36	12.2	Barley root	(29)
	3	1400	20.6		
	4	12000	46.8		
Na^+	1	44	7.3	Barley root	(30)
	2	1180	10.8		
	3	830	16.9		
	4	8400	35.7		
Mg^{2+}	1	20	1.35	Corn root	(31)
	2	120	2.41		
	3	1900	3.68		
Mn^{2+}	1	5.3	0.06	Citrus seedling	(32)
	2	7.2	0.19		
	3	64	1.63		
Cu^{2+}	1	9.3	1.5	Barley root	(33)
	2	300	36.5		
Cl^-	2	9.7	16.4	Barley root	(34)
	3	98	23.1		
	4	3000	41.2		
SO_4^{2-}	1	6.7	0.07	Barley root	(35)
	2	42	0.11		
	3	410	0.20		
HPO_4^{2-}	1	7.0	1.17	Barley root	(35)
	2	25	1.60		
	3	440	3.68		

of distinct kinetic phases; which based on corresponding K_m values indicates that the plant root possesses the affinity to absorb ions over a broad concentration range. For the data shown this represents a concentration range of three to four orders of magnitude. Secondly, a comparison of K_m values and potential uptake rates (V_{max}) indicate that there is a degree of control exerted in ion uptake which is related to the concentration of ions in soil solution. In effect, uptake is much more efficient at lower soil concentrations than at higher concentrations, for example, in the case of Mg (phases 1 and 2) a five fold increase in substrate ions results in less than a doubling of uptake rate. Finally, the consistency of this kinetic relationship for all nutrient ions studied, suggests that nonnutrient metals may behave similarly.

Although uptake mechanisms can be shown to be quite specific for individual ions, competition with respect to absorption can be shown for groups of closely related anions and cations. Vange et al. (35) have shown that sulfite, thiosulfate, molybdate, selenate, and chromate competitively inhibit sulfate uptake and therefore behave as analogs of sulfate, while divalent phosphate, perchlorate and periodate do not affect sulfate transport. Similar relationships have been shown for potassium-rubidium (36) and copper-zinc (37, 38). Since higher plants are known

to accumulate nonessential elements in appreciable quantities (39); these elements may be accumulated by mechanisms in place for absorption of chemically similar nutrilites. Unfortunately, little effort has been expended to understand the mechanisms regulating metal uptake by plants.

Since it can be readily shown that nonnutrient elements are accumulated by plants, what are the controlling mechanisms? Cutler and Rains (40) investigated the mechanisms of Cd uptake in excised roots of barley and concluded that absorption was nonmetabolic with uptake being the result of diffusion coupled to sequestration. However, these studies have several shortcomings. The Cd concentrations employed ranged from 1 to 20 ppm in solution, which is two to three orders of magnitude higher than would be encountered in soil solution, and well beyond physiological concentrations, especially if Cd were behaving as an analog of a required trace element. At high solution concentration of Cd, sorption may far outweigh the fraction of Cd being absorbed into the symplast. Finally, high Cd levels, especially with excised roots, may further have an inhibiting effect on metabolism and therefore permeability.

Although the work of Cutler and Rains (40) indicated that Cd uptake by plants was not metabolically regulated, the work of Vange et al. (35) suggests, as discussed earlier, that there are specific interactions in the absorption of nutrient and non-nutrient anions. Studies of the absorption of non-nutrient elements by the authors and coworkers have concentrated on Cd, Tl, and Ni. Since the absorption of micronutrients by plants saturates carrier systems at $\sim$200 μM, the kinetic studies were performed below this concentration. Many of the problems associated with Cd absorption (40) were resolved when Cd concentrations were limited to <1.0 μM in nutrient solutions. Examination of the relationship between Cd uptake by 18-day old intact soybean plants and Cd concentration (Fig. 1A) indicated that there were two distinct Cd absorption phases over the concentration of 0.01–0.40 μM. At Cd concentrations over 1.0 μM, reproducible kinetic relationships were not obtained, partly due to difficulties in removing adsorbed Cd from root surfaces, and obvious toxicity symptoms such as loss of turgor in shoots. The presence of multiple absorption isotherms with distinct transition is characteristic of ion uptake in higher plants, and suggests that Cd uptake may be under metabolic regulation. Analyses of these data using double reciprocal plots showed both absorption phases 1 and 2 to conform to Michaelis-Menten kinetics (Table 4). This indicates the involvement of a rate-limiting membrane transport step during Cd absorption.
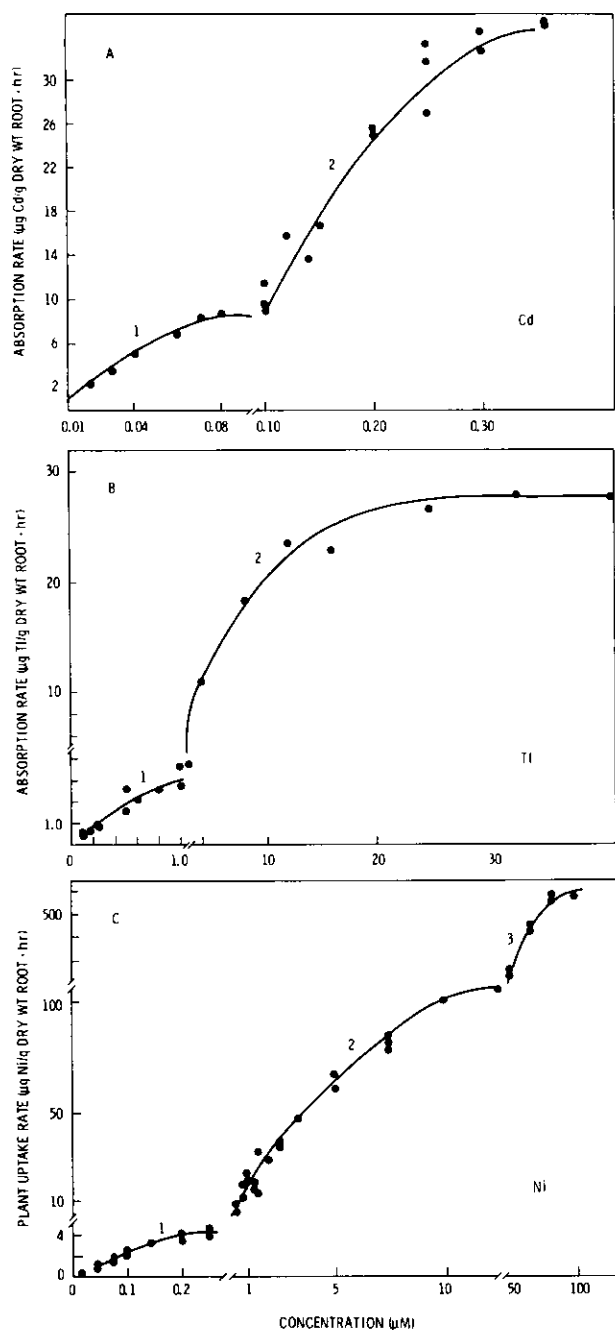
FIGURE 1. Relationship between metal uptake and concentration by intact soybean plants.

Similar concentration-dependent uptake relationships were obtained for Tl and Ni (Fig. 1B and 1C). These exhibited two- and three-phase transitions, respectively. Replots of the individual isotherms shows each to conform to Michaelis-Menten kinetics as in the case of Cd (Table 4). These kinetic constants, especially in the case of Ni, indicate that the plant root possesses effective and efficient mechanisms for accumulating nonnutrient ions over a broad range of metal concentration in soil solution as evident for nutrient ions (Table 3) and suggests that similar mechanisms may be in place for uptake of other heavy metals. The saturation behavior of ion absorption processes is important to the use of plants as monitor systems since the concentration of a pollutant in soil solution is not directly related to plant content. In addition, the multiphasic behavior results in nonlinearity of response between soil solution concentration of an ion and plant absorption.

Since the absorption of Cd, Tl, and Ni appears to be under metabolic regulation, ion competition studies were employed to determine if the absorption of these nonnutrilites resulted from behavior as analogs of essential nutrients. The interactions of a number of nutrient ions with Cd, Tl and Ni have been determined (Table 5). The absorption of Cd from 0.1 μM solutions was reduced by 17–33% in the presence of 0.5 μM Fe, Mn, Zn, and Cu. Detailed kinetic analyses of the mechanism of inhibition failed to resolve the nature of the interactions

Table 4. Kinetic constants for Cd, Tl, and Ni by using 18-21 day old intact soybean plants.

Element	Absorption phase	K_m, μM	V_{max}, mg/g dry wt root-hr	r^2
Cd	1	0.22	28	0.91
	2	1.67	200	0.86
Tl	1	1.82	8.3	0.95
	2	13.3	45	0.97
Ni	1	0.51	13	0.93
	2	8.6	175	0.94
	3	379	1870	0.96

Table 5. Interaction of nutrient ions with Cd, Tl, and Ni.[a]

Heavy metal	Competing coions	Inhibition of heavy metal uptake, %	Type of interaction[b]
Cd^{2+}	Zn^{2+}	25	Unknown (competitive)
	Cu^{2+}	33	Unknown (competitive)
	Fe^{2+}	17	Unknown
	Mn^{2+}	25	Unknown
Tl^+	K^+	57	Noncompetitive
Ni^{2+}	Cu^{2+}	42	Competitive
	Zn^{2+}	30	Competitive
	Fe^{2+}	33	Unknown
	Co^{2+}	25	Unknown

[a] Coions supplied in 5× excess of heavy metal concentration; Cd, Tl and Ni supplied at 0.1, 10, and 1.0 μM, respectively.
[b] Mechanism of interaction determined by using Lineweaver-Burke plots.

between Cd, Fe, and Mn but Zn and Cu do appear to be competitive inhibitors of Cd transport. Experimental variability due to physical adsorption of Cd prevented calculation of inhibitor constants (K_i). The Tl absorption was inhibited by 57% in the presence of a 10-fold higher concentrations of K. Detailed kinetic analysis of the interactions using concentrations within phase 1 (Table 5) demonstrated that K was a noncompetitive inhibitor of Tl transport. The calculated K_i value of 23 μM for classical noncompetitive inhibition indicates that a concentration of 23 μM K will reduce Tl absorption by 50% at all concentrations within the concentrations bracketed by phase 1. Although not determined, a similar situation can be expected to occur for phase 2 at its K_i value. The absorption of Ni was inhibited by 25–42% in the presence of Cu, Co, Fe, and Zn. Detailed kinetic analyses failed to resolve the interaction of Co and Fe with Ni; however, both Cu and Zn were shown to be competitive inhibitors of Ni transport. The K_m and K_i constants for Ni in the absence and presence of Cu were calculated to be 6.1 and 0.2 μM, respectively, while K_m and K_i constants for Ni in the absence and presence of Zn were 6.7 and 24.4 μM, respectively. Since the K_i values for Cu and Zn represent the inhibitor concentration necessary to double the slope of the $1/v$ versus $1/[S]$ plot, Cu is shown to be a better competitor of Ni and Zn. The affinity of these carrier sites for Cu is about 60% of that for Ni, while their affinity for Zn is only 25% that of Ni. The kinetic parameters indicate that at a constant soil solution concentration of Cu and/or Zn, Ni uptake by plants will decrease as the Ni concentration is reduced through the concentration range comprising phase 2 (Fig. 1C).

A kinetic approach to the behavior of heavy metal in soils and plants, aside from providing an insight into controlling mechanisms, uptake potential, and ion associations, is useful in interpreting and identifying some of the current problems associated with environmental dissemination of heavy metals from anthropogenic activities. For example, it is clear that the use of municipal sludge amendments for agricultural soils will result in higher plant concentrations of several heavy metals, but, in addition, amendments may also reduce the uptake of required nutrient analogs; similarly, the concentration of nutrient analogs in soil solution will affect the rate of absorption on nonnutrients.

Metabolic Behavior of Metals in Plants as Related to Tolerance

The feasibility of employing higher plants as monitors of metal pollution is dependent upon an understanding of the metabolic processes which en-

able plants to acquire needed nutrients and tolerate increasing levels of toxic elements. Plant breeding studies have led to the realization that many aspects of mineral nutrition are under genetic regulation and therefore governed by selection. Genetic control has been shown to govern the initial absorption of ions (41, 42) the oxidation-reduction of Fe (43), compartmentalization of ions within the root (44), transfer from root to xylem (45), and metabolic utilization (46, 47). Similarly, tolerance of plants to high levels of both nutrient and nonnutrient elements appears to be genetically controlled (48, 49).

Tolerance of plants to individual metals, although forming the functional basis for the behavior of "indicator" or "accumulator" plants, is often misconceived. In the case of accumulator species, their adaptation of geographical areas containing high concentrations of endogenous metals involved genotypic evolution and selection over a period of time induced by the specific habitat. Man-made alterations in the environment, such as those resulting from mining operations, involve similar selection pressures, as is evident from cursory examination of these sites. Although the number of ecotypes is limited, the process of adaptation is relatively rapid. This would suggest that the indigenous wild population contains sufficient genetic variability to produce individuals capable of withstanding these adverse conditions.

Apparent plant tolerance can result from exclusion of toxic elements or metabolic tolerance to specific elements; the latter appears to be the more prevalent mechanism. Mechanisms for exclusion have been shown to include low root cation exchange capacities limiting uptake of Al and Mn (49), sorption of Zn to cell walls (50), and precipitation of Al by hydroxyl ions at the root surface (51, 52).

Metabolic adaptation as a mechanism of metal tolerance in plants appears to be the rule rather than the exception. A comparison of individual grass species (Agrostis tenuis) collected from old mine dumps and adjacent pasture lands and stressed under laboratory conditions, resulted in identification of important traits of tolerant species (53). Plant populations derived from environments containing elevated levels of lead were tolerant to lead but not other metals; similar results were obtained for plants tolerant to other metals, suggesting that tolerance is specific. The observed tolerance of adapted plants and the sensitivities of plants growing in adjacent areas of low metal burden were not lost on cultivation although individuals did differ, and seeds collected from the tolerant population had the same tolerances as the parent and vegetatively propagated plants.

What metabolic changes in tolerant species could

account for reduced metal toxicity? The major mechanisms appear to be compartmentalization, complexation and metabolic adaptation. Compartmentalization would provide a means of limiting the presence of toxic metals at cellular locations where toxicity is initiated. For example, Al effects are most pronounced on processes of cell division and respiration; and compartmentalization or exclusion from these metabolic sites, provides, an effective protective mechanism (54). A plant adaptation which may play a role to tolerance is the alteration of metabolic sequences to allow an organism to function in an apparently normal manner in the presence of large amounts of heavy metals (55). To date, modification in metabolic pathways has been demonstrated in Cu tolerant yeast and bacteria, but this mechanism has not yet been reported to occur in higher plants.

Complexation may also serve to alleviate the toxicity of heavy metals in plants. It appears to be a common process in plants, and therefore may also represent a common mechanism for tolerance to toxic metals. Investigations of the forms of nutrient cations such as Fe, Cu, and Zn in leaf tissues of agronomic species (56, 57) have shown these to be present as anionic complexes. Similarly, studies of the chemical forms of trace elements transported within the xylem (11, 25, 37–39, 58–62) and phloem (63) show Ca, Fe, Ni, Mn, and Zn to be transported as organic complexes. In the case of nutrient ions, complexation may provide a basis for maintaining their mobility within the plant and allowing for their accumulation at sites of metabolic use. A number of studies have dealt with the chemical form of Ni in leaves of accumulator species (64–66) and agricultural plants (67, 68). These have shown Ni to exist in low molecular weight cationic and anionic complexes, and therefore, may represent an example of the role of complexation in tolerance.

Plants as Monitors of Metal Pollution

In order for plants to serve as indicators of incremental increases of metals in soil, deposition must be sufficient to significantly increase the soluble, and ultimately, the plant-available, fraction in soil. Furthermore, increases must be distinguishable from changes in the soluble fraction which occur with time and weathering of minerals. These requirements are complicated by the broad range in total metal concentrations and form in soil and the environmental conditions under which the soils exist.

Studies to date indicate that increased soil levels resulting from diffuse sources such as stack emissions from coal combustion may not be detectable by measuring plant uptake and effects. Increases in soil–metal levels may be significant in cases of localized pollution such as in disposal of bottom ash and scrubber wastes from fossil fuel combustion, industrial wastes and municipal sewage sludges resulting in detectable increases in plants. However, even in situations of significant increases in metal levels, the wastes may, in themselves, modify soil properties influencing metal solubility and the value of plants as monitors of pollutant levels will be dependent on a detailed understanding and precise measurements of influential soil and plant variables. In the case of soil there is a need to understand those factors influencing solubility and form of the metals with times. In the case of plants, information is required regarding physical processes, membrane transport (ion interactions, kinetics) and metabolic fate, particularly as influenced by metal stress.

Membrane transport will result in increased levels of metals within the plant based on available concentrations in soil solution. This can lead to a 10–1000× concentration by plants, at least for nutrient species, improving the feasibility for the use of the plant for detection of changes in environmental levels. On the basis of Cd, Tl, and Ni behavior, it would appear that metals may exhibit similar tendencies. However, as demonstrated by ion competition studies, the uptake of these nonnutrient species will not necessarily be proportional to soil solution concentration, particularly at environmental concentration levels, and will be attenuated by the presence and concentration of their respective nutrient analogs. Once absorbed, the metabolic fate of metals will determine, not only their partitioning between root and shoot, but their toxicity. Although partitioning will undoubtedly vary with the element, toxicity should not be a major consideration at most environmental concentrations.

In summary, for specific sites with local pollution sources, plants may be useful monitors of increases in metals, but this will require a specific understanding of soil solubilization and plant uptake mechanisms.

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